

What is claimed is:

1. A device (1) for measuring the lifetime of the fluorescence of fluorophores in samples, the device including at least one light source (2) for exciting  
5 the fluorescence of the fluorophores, an irradiation optic (3) for directing the excitation light (4) onto these samples, a sample table (5) for placing a microplate (6), which contains samples, at the irradiation optic (3), an emission optic (7) for directing the fluorescence light (8) from the samples onto a detector (9), and at least one detector (9) having analysis electronics,  
10 wherein the irradiation optic (3) of the device (1) includes a beam splitter (10) having at least two mirrors (11), which directs a part of the light (4) from the at least one light source (2), which always enters the beam splitter (10) with the same power and the same pulse shape along a first optical axis (12), in the direction of a sample and allows a part of this light to  
15 pass on to the respective mirror (11) lying behind it.
2. The device (1) according to Claim 1,  
wherein the beam splitter (10) is implemented as a mirror slide (13),  
20 movable along this first optical axis (12), and it deflects the light (4) from the first light source (2) by 90° in the direction of a sample.
3. The device (1) according to Claim 1,  
wherein the mirrors (11) of the beam splitter (10) allow 75% of the incident light (4) to reflect and 25% of this light to pass onto the respective  
25 mirror (11) lying behind it.
4. The device (1) according to Claim 1, which also includes a further light source (18),  
30 wherein the beam splitter (10) is implemented so it may be removed from the device (1) and replaced by a filter element (19), the filter element (19) including at least one filter (21), which allows a part of the light of the light source (18) to pass in the direction of the sample.

5. The device (1) according to Claim 4,  
wherein the filter element (19) is implemented as a filter slide (20), movable along this first optical axis (12), the bandpass of the filters (21) being identical or different.
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6. The device (1) according to Claim 2,  
wherein the mirror slide (13) or filter slide (20) is implemented so it may be automatically moved, using a drive, into the correct position for a specific sample.
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7. The device (1) according Claim 1, the device (1) including two light sources (2) in the form of lasers,  
wherein the two lasers are coupled to a first optical waveguide (25) and/or second optical waveguide (25') and the two optical waveguides (25, 25') feed the light of the two lasers to a connection point in a collecting lens (22), using which the two focal points thus defined may be positioned in a specific well of a microplate (6).
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8. The device (1) according to Claim 7,  
in which, at the connection point (22) in the region of the end (29) of the first and second optical waveguides (25, 25'), the sheathing (27, 27') of these optical waveguides is removed and replaced by a shared sheathing (30), so that the optical axes (23, 23') of the two optical waveguides (25, 25') lie at a minimum distance (A) to one another.
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9. The device (1) according to Claim 8,  
wherein the minimum distance (A) is 125  $\mu\text{m}$ .
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10. The device according to Claim 1,  
wherein the device includes a processor for controlling the device (1) and for automatic analysis of the measurement data of the detectors (9, 9', 9'') and a drive of the sample table (5) for automatic positioning of the wells of a microplate (6) at the irradiation optic (3) and emission optic (7).
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11. A method of measuring the lifetime of the fluorescence of fluorophores in samples in a device (1), the device including at least one light source (2) for exciting the fluorescence of the fluorophores, an irradiation optic (3) for directing the excitation light (4) onto these samples, a sample table (5) for placing a microplate (6), which contains a sample, at the irradiation optic (3), an emission optic (7) for directing the fluorescence light (8) from the samples onto a detector (9), and at least one detector (9) having analysis electronics,
- wherein the light from the at least one light source (2) is always fed with the same power and the same pulse shape along a first optical axis (12) into the beam splitter (10), the beam splitter (10) including at least two mirrors (11), using which a part of the light (4) from the at least one light source (2) is deflected in the direction of a sample and a part of this light (4) is passed through to the respective mirror (11) lying behind it.
12. The method according to Claim 11,
- wherein the beam splitter (10) is implemented as a mirror slide (13) movable along this first optical axis (12), the mirrors (11) of the beam splitter (10) each allowing 75% of the incident light to reflect by 90° in the direction of a sample and 25% of this light to pass onto the respective mirror (11) lying behind it.
13. The method according to Claim 11, in which a device (1) is used which also includes a further light source (18),
- wherein the beam splitter (10) is removed from the device (1) and replaced by a filter element (19), the filter element (19) being implemented as a filter slide (20), movable along this first optical axis (12), having one or more filters (21) - which allow a part of the light of the further light source (18) to pass in the direction of the sample.

14. The method according to Claim 12,  
wherein the mirror slide (13) and/or the filter slide (20) is/are automatically moved using a drive into the correct position for a specific sample.
- 5 15. The method according to Claim 11, a device (1) being used which includes  
two light sources (2) in the form of lasers,  
wherein the two lasers are coupled to a first optical waveguide (25) and  
second optical waveguide (25'), respectively, and the two optical wave-  
guides (25, 25') feed the light of the two lasers to a connection point in a  
10 collecting lens (22), which positions the two focal points thus defined in a  
specific well of a microplate (6).
16. The method according to Claim 11,  
which has the following operating steps:
- 15 1) Performance of a reference measurement without the presence of a  
sample to determine the device constants.
- 2) Establishing the boundary conditions for the correlation parameters  
starting from the reference measurement:
- a) Determining time (33) at which maximum (31) occurs;
- 20 b) Determining time (36) at which the adjustment ends, a constant  
time interval (39) being subtracted of the end of time  
window (36);
- 3) Measuring the samples in the wells of a microplate.
- 25 17. The method according to Claim 16,  
in which, during measurement of the samples in the wells of a microplate  
(6), an adjustment of the sample data is performed which includes the following  
steps:
- a) Biexponential pre-adjustment using the boundary conditions from  
30 2), the results are two lifetimes;
- b1) If the first lifetime is short and the first amplitude associated  
therewith represents a significant proportion of the total amplitude,

the adjustment is started beginning from time (33) plus a short time interval (38);

b2) If the conditions of b1) are not fulfilled, the adjustment is started beginning from time (33) plus a long time interval;

5 c1) If the second lifetime is much longer than the first lifetime, a biexponential adjustment is performed, the two lifetimes being determined;

c2) If the conditions of c1) are not fulfilled, a monoexponential adjustment is performed, the single lifetime resulting.

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18. A computer program product for controlling the device (1) and for automatic analysis of the measurement data,  
wherein a computer program activated in a computer allows a processor, using the device (1) - according to at least one of Claims 1 to 10 - to approach at least one well of a microplate (6), to activate the probe in the well using excitation light (4), to measure the lifetime of the fluorescence (8) emitted by the sample, and to assign the sample a classification number which characterizes this lifetime.

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20 19. A computer program product for controlling the device (1) and for automatic analysis of the measurement data,  
wherein a computer program activated in a computer allows a processor, using the method according to at least one of Claims 11 to 17, to approach at least one well of a microplate (6), to activate the probe in the well using excitation light (4), to measure the lifetime of the fluorescence (8) emitted by the sample, and to assign the sample a classification number which characterizes this lifetime.

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